CLAIMS

We claim:

1. A method of cloning a target nucleic acid comprising:
   a) providing an enhanced homologous recombination (EHR) composition comprising:
      i) a recombinase;
      ii) a first and a second targeting polynucleotide, wherein said first polynucleotide comprises a fragment of said target nucleic acid and is substantially complementary to said second target polynucleotide;
      and iii) a separation moiety;
   b) contacting said EHR composition with a target library under conditions wherein said targeting polynucleotides can hybridize to said target nucleic acid; and
   c) isolating said target nucleic acid; wherein said providing and contacting are done using a robotic system.

2. The method according to claim 1 wherein said target nucleic acid is a target gene.

3. The method according to claim 2 wherein said target nucleic acid is a portion of said target gene.

4. The method according to claim 1 wherein said target nucleic acid is a regulatory sequence.

5. The method according to claim 1 further comprising:
   d) making a library of nucleic acid variants of said target nucleic acid;
   e) introducing said library of nucleic acid variants into a target library; and
   f) performing phenotypic screening on said cellular library.

6. The method according to claim 1 wherein at least one of said making, introducing and performing steps are done using a robotic system.

7. The method according to claim 1 further comprising:
   d) making a plurality of cells comprising a mutant target nucleic acid;
   e) adding a library of candidate agents to said plurality;
   f) determining the effect of said candidate agents on said cells; and
   g)determining the effect of said candidate agent on said gene products.

8. The method according to claim 7 wherein at least one of said making, adding, and determining steps are done using a robotic system.

9. The method according to claim 7 wherein said mutant target nucleic acid is a gene sequence knock-out or a gene sequence knock-in.

10. The method according to claim 7 wherein said mutant target nucleic acid comprises an insertion, substitution, deletion or combinations thereof.

11. The method according to claim 1, wherein said robotic system comprises a computer workstation comprising a microprocessor programmed to manipulate a device selected from the group consisting of a thermocycler, a multichannel pipettor, a sample handler, a plate handler, a gel loading system, an automated transformation system, a gene sequencer, a colony picker, a bead picker, a cell sorter, an incubator, a light microscope, a fluorescence microscope, a spectrofluorimeter, a spectrophotometer, a luminometer a CCD camera and combinations thereof.

12. ~~13.~~ A method of high throughput integrated genomics comprising:

a) providing a plurality of enhanced homologous recombination (EHR) compositions, wherein each composition comprises:

i) a recombinase;

ii) a first and a second targeting polynucleotide, wherein said first polynucleotide comprises a fragment of said target nucleic acid and is substantially complementary to said second target polynucleotide;

and iii) a separation moiety;

b) contacting said EHR compositions with one or more nucleic acid sample(s) under conditions wherein said targeting polynucleotides hybridize to one or more target nucleic acid member(s) of said one or more libraries; and c) isolating said target nucleic acid(s); wherein said providing and contacting are done using a robotic system.

13. ~~14.~~ The method according to claim 13 wherein said target nucleic acid is a target gene.

14. ~~15.~~ The method according to claim 14 wherein said target nucleic acid is a portion of said target gene.

15. ~~16.~~ The method according to claim 13 wherein said target nucleic acid is a regulatory sequence.

16. ~~17.~~ The method according to claim 13 wherein said isolated target nucleic acids comprise single-nucleotide polymorphisms, a gene family, a haplotype.

17. ~~18.~~ The method of claim 13 wherein said nucleic acid sample(s) are selected from the group consisting of a cDNA library, genomic DNA library, genomic DNA samples, and combinations thereof.

18. ~~19.~~ The method of claim 18 wherein said genomic DNA samples are from one or more organisms or

patients.

20 ~~20~~. The method according to claim 13 further comprising:

d) making a library of nucleic acid variants of said target nucleic acid;

e) introducing said library of nucleic acid variants into a cellular library; and

f) performing phenotypic screening on said cellular library.

20 ~~21~~. The method according to claim 20 wherein at least one of said making, introducing and performing steps are done using a robotic system.

21 ~~22~~. The method according to claim 13 further comprising:

d) making a plurality of cells comprising a mutant target nucleic acid;

e) adding a library of candidate agents to said plurality; and

f) determining the effect of said candidate agents on said cells.

22 ~~23~~. The method according to claim 22 wherein at least one of said making, adding, and determining steps are done using a robotic system.

23 ~~24~~. The method according to claim 22 wherein said mutant target nucleic acid is a gene sequence knock-out or a gene sequence knock-in.

24 ~~25~~. The method according to claim 22 wherein said mutant target nucleic acid comprises an insertion, substitution, deletion or combinations thereof.

25 ~~26~~. The method of claim 13 further comprising;

d) introducing said target nucleic acid(s) into one or more cell(s), wherein said introducing is done using a robotic system.

26 ~~27~~. The method of claim 26 further comprising;

e) expressing said target nucleic acid(s), wherein said expressing is done using a robotic system.

27 ~~28~~. The method of claim 27 further comprising;

f) identifying a cell(s), embryo(s), organism(s) having an altered phenotype induced by a biological activity of the expressed target nucleic acid, wherein said identifying is done using a robotic system.

28 ~~29~~. The method according to claim 27, further comprising sequence said expressed target nucleic acid.

29 30. The method according to claim 27, further comprising mapping said expressed target nucleic acid.

Rule 1.126 30 31. The method according to claim 27, wherein said altered phenotype comprises altered expression of a cellular gene.

31 32. The method of claim 28 further comprising;

g) contacting said cell(s) having an altered phenotype with a library of candidate bioactive agents, wherein said contacting is done using a robotic system.

32 33. The method of claim 32 further comprising;

h) identifying a bioactive agent that modulates an activity of the expressed target nucleic acid, wherein said identifying is done using a robotic system.

33 34. The method of claim 13, 21, 23, 26, 27, 28, 32 or 33 wherein said robotic system comprises a computer workstation comprising a microprocessor programmed to manipulate a device selected from the group consisting of a thermocycler, a multichannel pipettor, a sample handler, a plate handler, a gel loading system, a gene sequencer, an automated transformation system, a colony picker, a bead picker, a cell sorter, an incubator, a light microscope, a fluorescence microscope, a spectrofluorimeter, a spectrophotometer, a luminometer a CCD camera and combinations thereof.

34 35. A robotic system comprising:

a) means for producing a plurality of enhanced homologous recombination compositions.

35 36. The system of claim 35 further comprising:

b) means for contacting said compositions with a cellular library under conditions wherein said compositions hybridize to one or more target nucleic acid members of said library.

36 37. The system of claim 36 further comprising:

c) means for isolating said target nucleic acid(s).

37 38. The system of claim 37 further comprising a means for producing a library of mutant target nucleic acid(s).

38 39. The system of claim 37 further comprising a means for nucleotide sequencing said target nucleic acid(s).

39 40. The system of Claim 37 further comprising a means for determining the haplotype of said target nucleic acid.

40 ~~41~~. The system of claim 40 further comprising:

Rule 1.126

d) means for introducing said target nucleic acid(s) into host cells.

41 ~~42~~. The system of claim 41 further comprising:

e) means for expressing said target nucleic acid(s) in said cells.

42 ~~43~~. The system of claim 42 further comprising:

f) means for identifying one or more cell(s) having an altered phenotype induced by a biological activity of said expressed target nucleic acid(s).

43 ~~44~~. The system of claim 43 further comprising:

g) means for contacting said cell(s) with a library of candidate bioactive agents.

44 ~~45~~. The system of claim 44 further comprising:

h) means for identifying one or more bioactive agent(s) that modulate a biological activity of said expressed target nucleic acid(s).

45 ~~46~~. The system of any one of claims 35-45 wherein said robotic system comprises a computer workstation comprising a microprocessor programmed to manipulate a device selected from the group consisting of a thermocycler, a multichannel pipettor, a sample handler, a plate handler, a gel loading system, an automated transformation system, a gene sequencer, a colony picker, a bead picker, a cell sorter, an incubator, a light microscope, a fluorescence microscope, a spectrofluorimeter, a spectrophotometer, a luminometer, a CCD camera and combinations thereof.